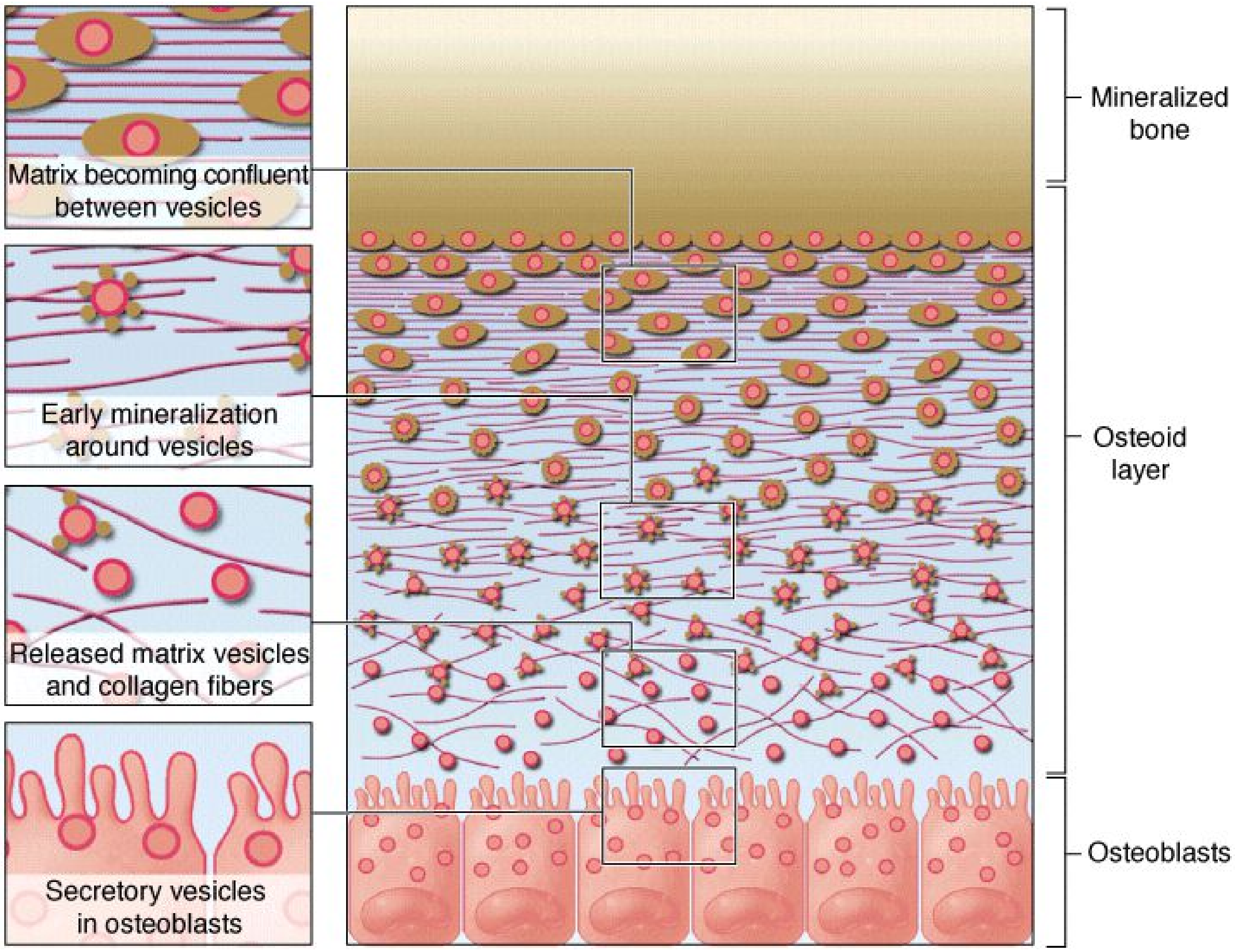
Bone Development

9th lecture

December 17, 2015

Mineralization in bone matrix (calcification)

- Osteoblasts secrete type I collagen, several glycoproteins, and proteoglycans.
- Some of these factors, notably osteocalcin and certain glycoproteins, bind Ca^{2+} with high affinity, thus raising the local concentration of these ions.
- Osteoblasts also release very small membrane-enclosed matrix vesicles with which alkaline phosphatase and other enzymes are associated.
- These enzymes hydrolyze PO_4 ions from various macromolecules, creating a high concentration of these ions locally.
- The high ion concentrations cause crystals of $CaPO_4$ to form on the matrix vesicles.
- The crystals grow and mineralize further with formation of small growing masses of hydroxyapatite [$Ca_{10}(PO_4)_6(OH)_2$] which surround the collagen fibers and all other macromolecules.
- Eventually the masses of hydroxyapatite merge as a confluent solid bony matrix as calcification of the matrix is completed.



Matrix becoming confluent between vesicles
Early mineralization around vesicles
Released matrix vesicles and collagen fibers
Secretory vesicles in osteoblasts
Mineralized bone
Osteoid layer
Osteoblasts

Osteogenesis

Bone can be formed initially by either of two ways:

- Endochondral ossification, in which the matrix of preexisting hyaline cartilage is eroded and replaced by osteoblasts producing osteoid.

- Intramembranous ossification, in which osteoblasts differentiate directly from mesenchyme and begin secreting osteoid

In both processes, the bone tissue that appears first is primary or woven. Primary bone is a temporary and is soon replaced by the definitive secondary lamellar bone. During bone growth, areas of primary bone, areas of resorption, and areas of secondary bone all appear side by side.

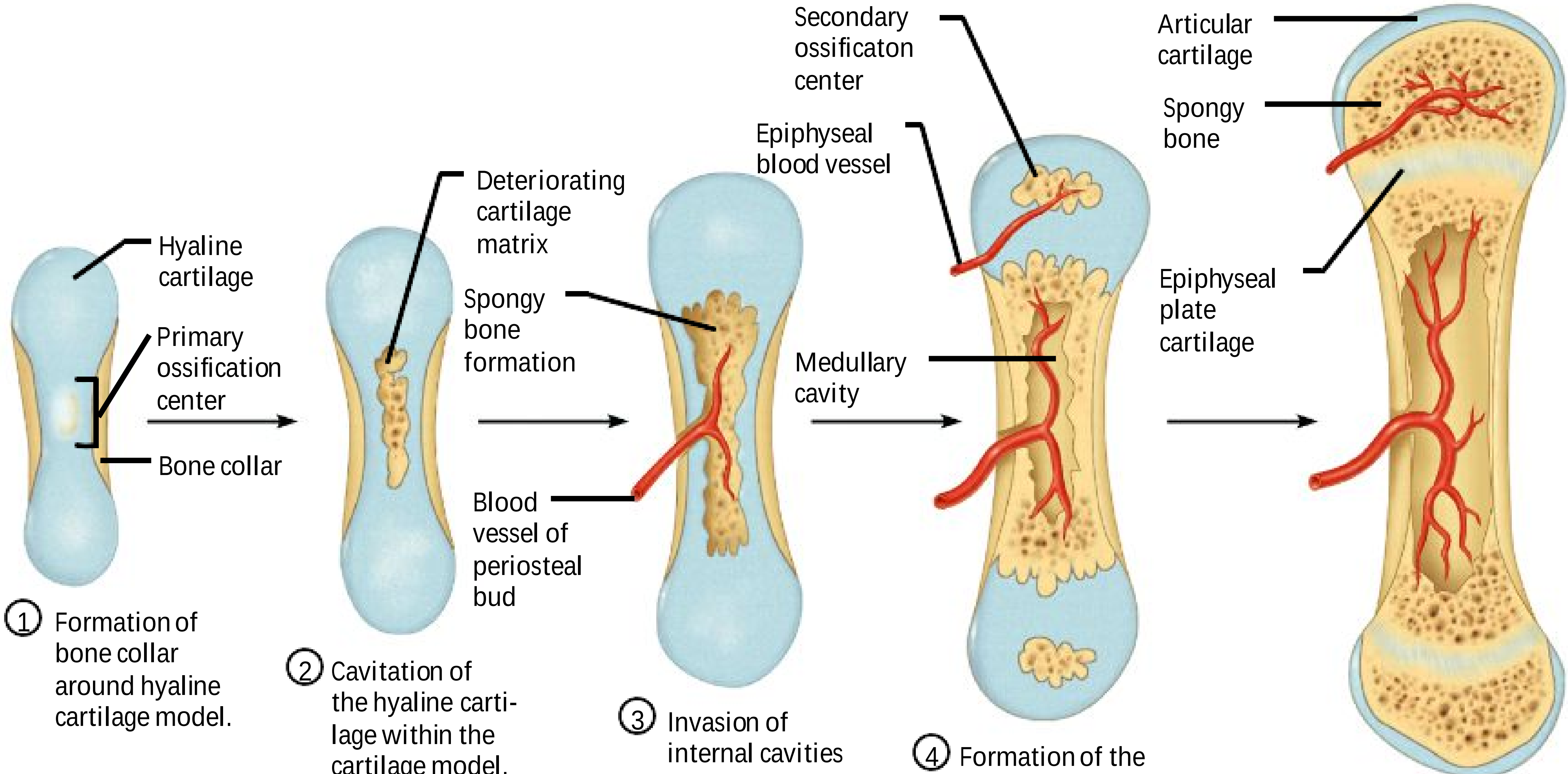
Endochondral ossification, forms most bones of the skeleton and occurs in the fetus in models made of hyaline cartilage

Stages of Endochondral Ossification

The process takes many weeks and major developmental stages include:

- (1), formation of a bone collar around the middle of the cartilage model and degeneration of the underlying cartilage
- (2), followed by invasion of the resulting ossification center by capillaries and osteoprogenitor cells from the periosteum
- (3), osteoid deposition by the new osteoblasts, calcification of woven bone, and its remodeling as compact bone
- (4), This primary ossification center develops in the diaphysis, along the middle of each developing bone. Secondary ossification centers develop somewhat later by a similar process in the epiphyses. The primary and secondary ossification centers are separated by the epiphyseal plate
- (5), which provides for continued bone elongation. The two ossification centers do not merge until the epiphyseal plate disappears
- (6), when full stature is achieved.

Stages of Endochondral Ossification

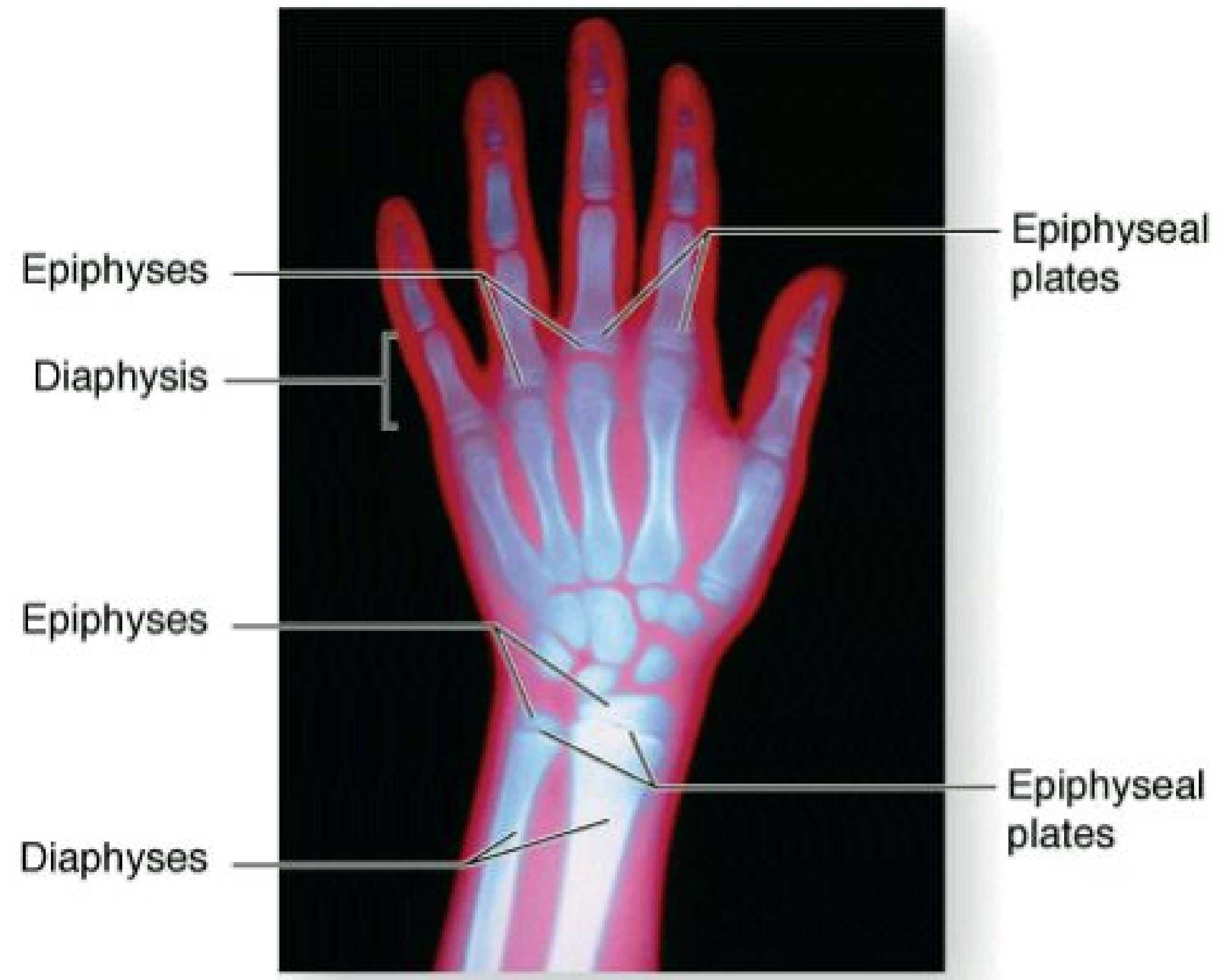


1. Formation of bone collar around hyaline cartilage model.
2. Cavitation of the hyaline cartilage within the cartilage model.
3. Invasion of internal cavities by the periosteal bud and spongy bone formation.
4. Formation of the medullary cavity as ossification continues; appearance of secondary ossification centers in the epiphyses in preparation for stage 5.
5. Ossification of the epiphyses; when completed, hyaline cartilage remains only in the epiphyseal plates and articular cartilages.

Epiphyseal growth plate: locations and zones of activity.

The large and growing primary ossification center in long bone diaphyses and the secondary ossification centers in epiphyses are separated in each developing bone by a plate of cartilage called the epiphyseal plate.

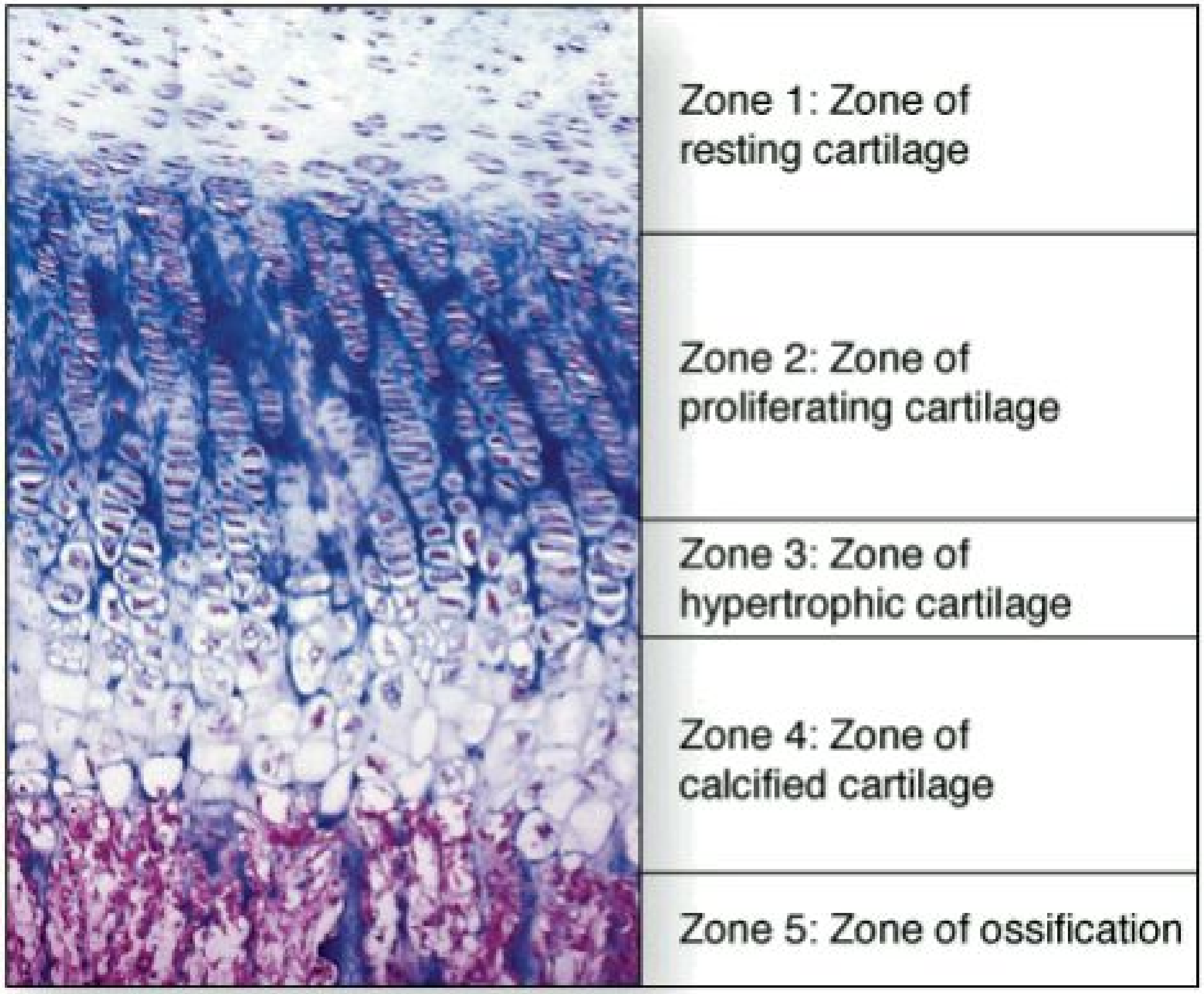
(a): Epiphyseal plates can be identified in an x-ray of a child's hand as marrow regions of lower density between the denser ossification centers. Cells in epiphyseal growth plates are responsible for continued elongation of bones until the body's full size is reached.



a X-ray of a hand

A plate of epiphyseal cartilage is divided into five zones, starting from the epiphyseal side of cartilage:

1. The resting zone consists of hyaline cartilage with typical chondrocytes.
2. In the proliferative zone, chondrocytes begin to divide rapidly and form columns of stacked cells parallel to the long axis of the bone.
3. The hypertrophic cartilage zone contains swollen chondrocytes whose cytoplasm has accumulated glycogen. Hypertrophy compresses the matrix into thin septa between the chondrocytes.
4. In the calcified cartilage zone, loss of the chondrocytes by apoptosis is accompanied by calcification of the septa of cartilage matrix by the formation of hydroxyapatite crystals.
5. In the ossification zone, bone tissue first appears. Capillaries and osteoprogenitor cells originating from the periosteum invade the cavities left by the chondrocytes. Many of these cavities will be merged and become the marrow cavity.

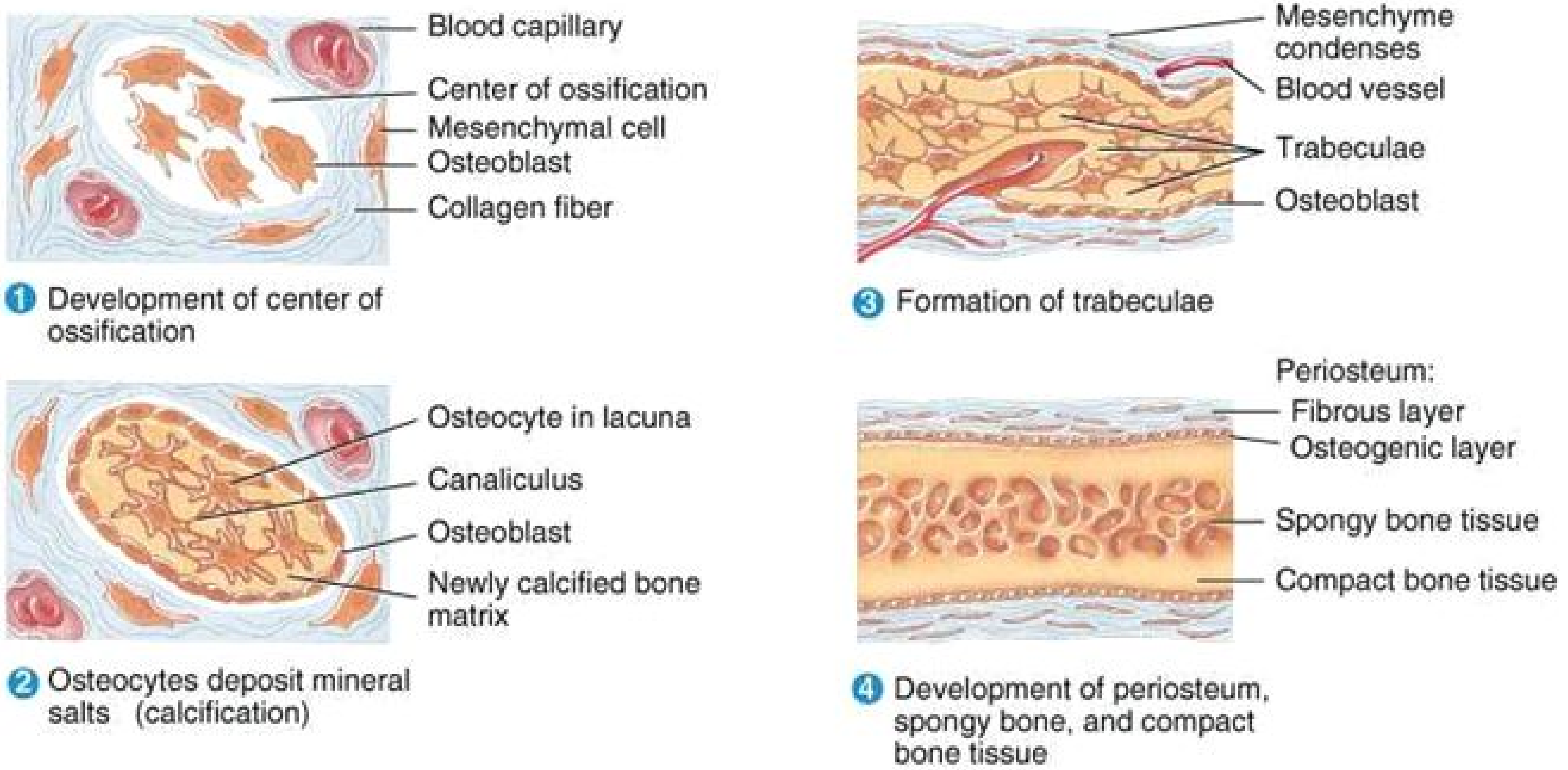


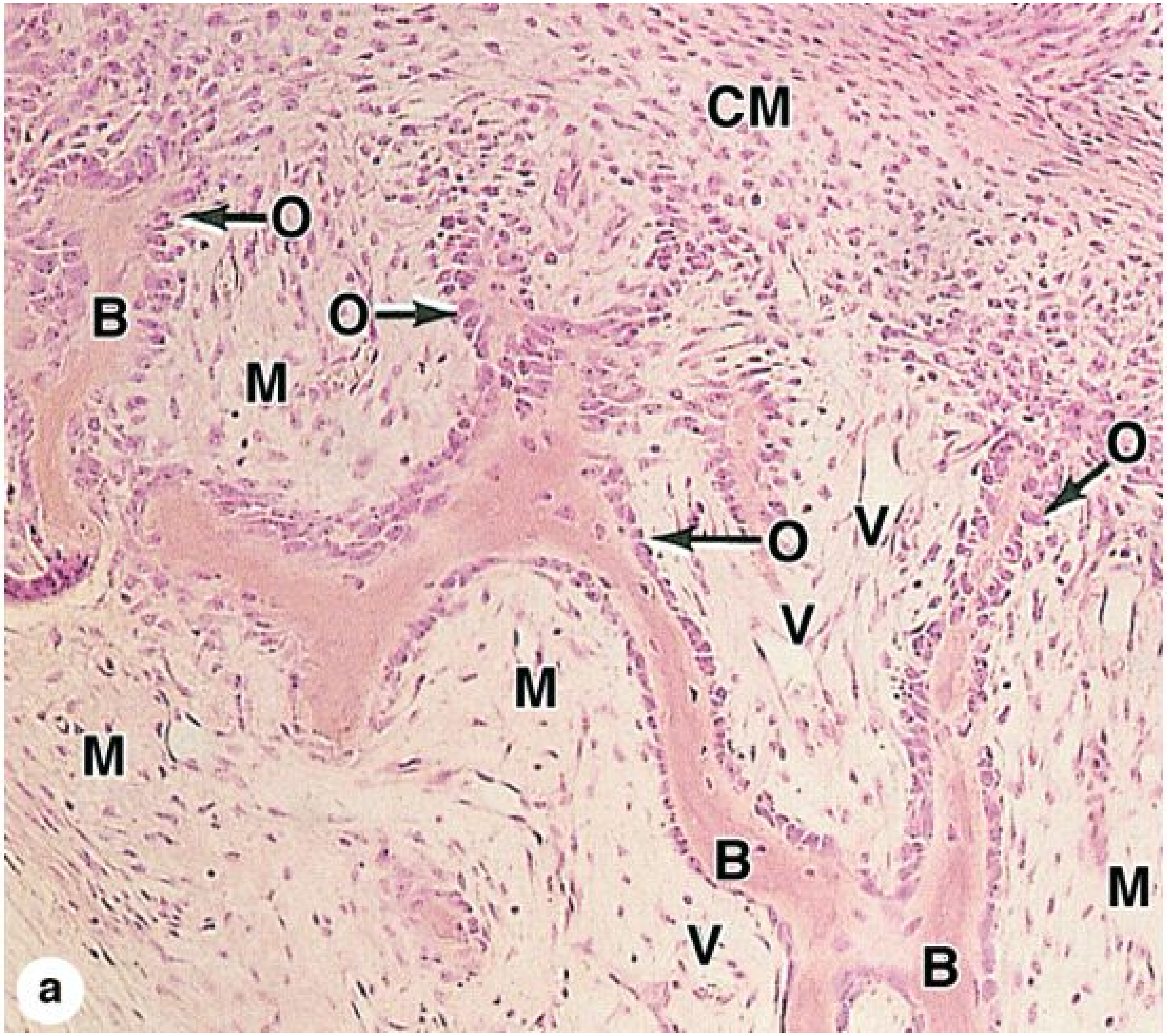
b Epiphyseal plate

Intramembranous Ossification

- Intramembranous ossification, by which most flat bones are produced
- is so called because it takes place within condensations of embryonic mesenchymal tissue.
- The frontal and parietal bones of the skull—as well as parts of the occipital and temporal bones and the mandible and maxilla—are formed by intramembranous ossification.

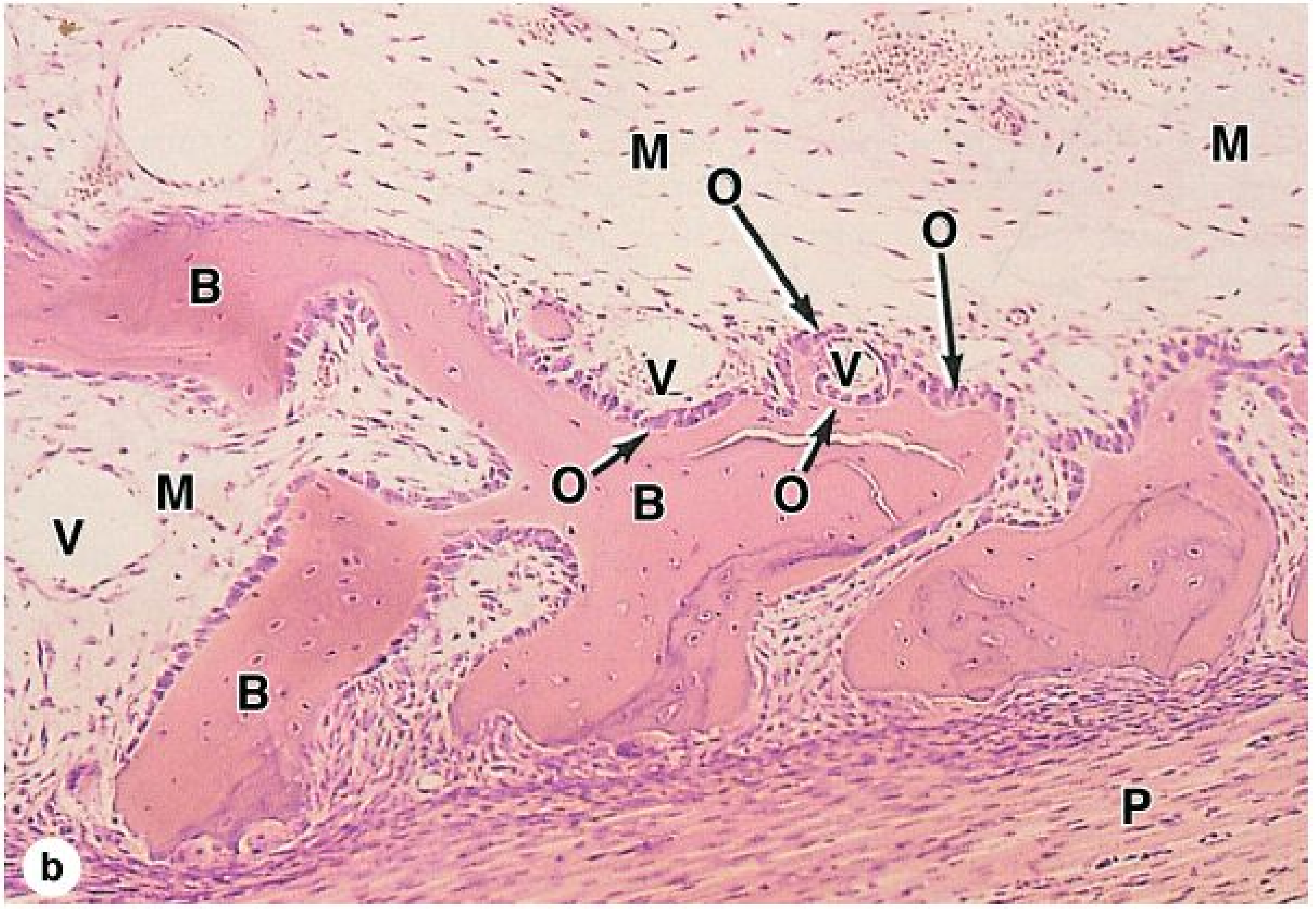
Intramembranous (between membranes) Ossification occurs in flat bones





Intramembranous ossification.

A section of jaw from a fetal pig undergoing intramembranous ossification. (a): Areas of typical mesenchyme (M), condensed mesenchyme (CM) adjacent to aggregates of new osteoblasts (O). Some osteoblasts have secreted matrices of bone (B) which remain covered by osteoblasts. Between these trabeculae of newly formed primary bone are vascularized areas (V) that will form marrow cavities. X40. H&E.



(b): Higher magnification shows the developing periosteum (P) that covers masses primary bone that will soon merge to form a continuous plate of bone. The larger mesenchyme-filled region at the top is the developing marrow cavity. X100. H&E.

Long Bone Growth and Remodeling

- Growth in length – cartilage continually grows and is replaced by bone as shown
- Remodeling – bone is resorbed and added by appositional growth as shown
  - compact bone thickens and strengthens long bones with layers of circumferential lamellae

Long Bone Growth and Remodeling

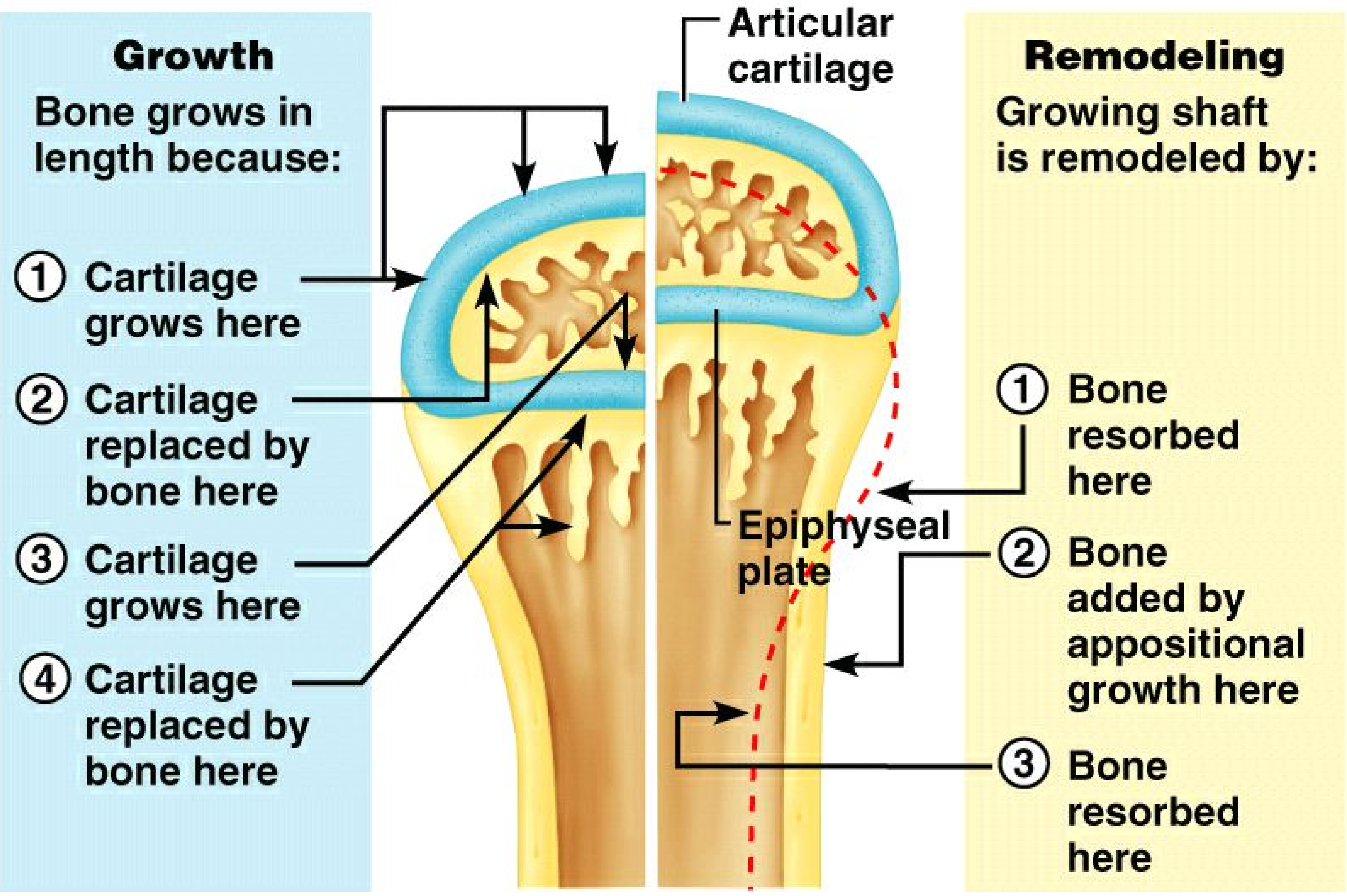
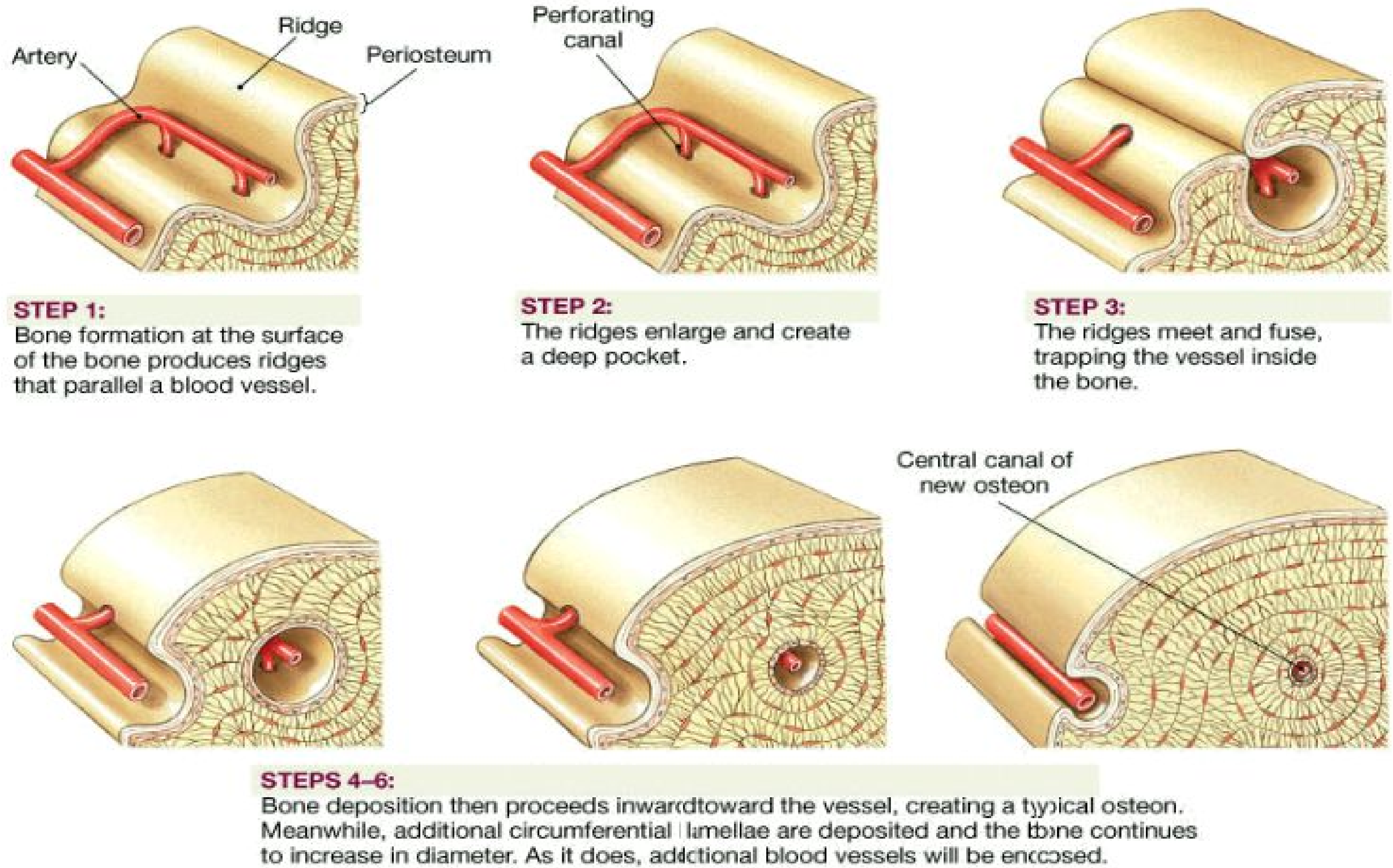


Figure 6.10

Appositional Growth



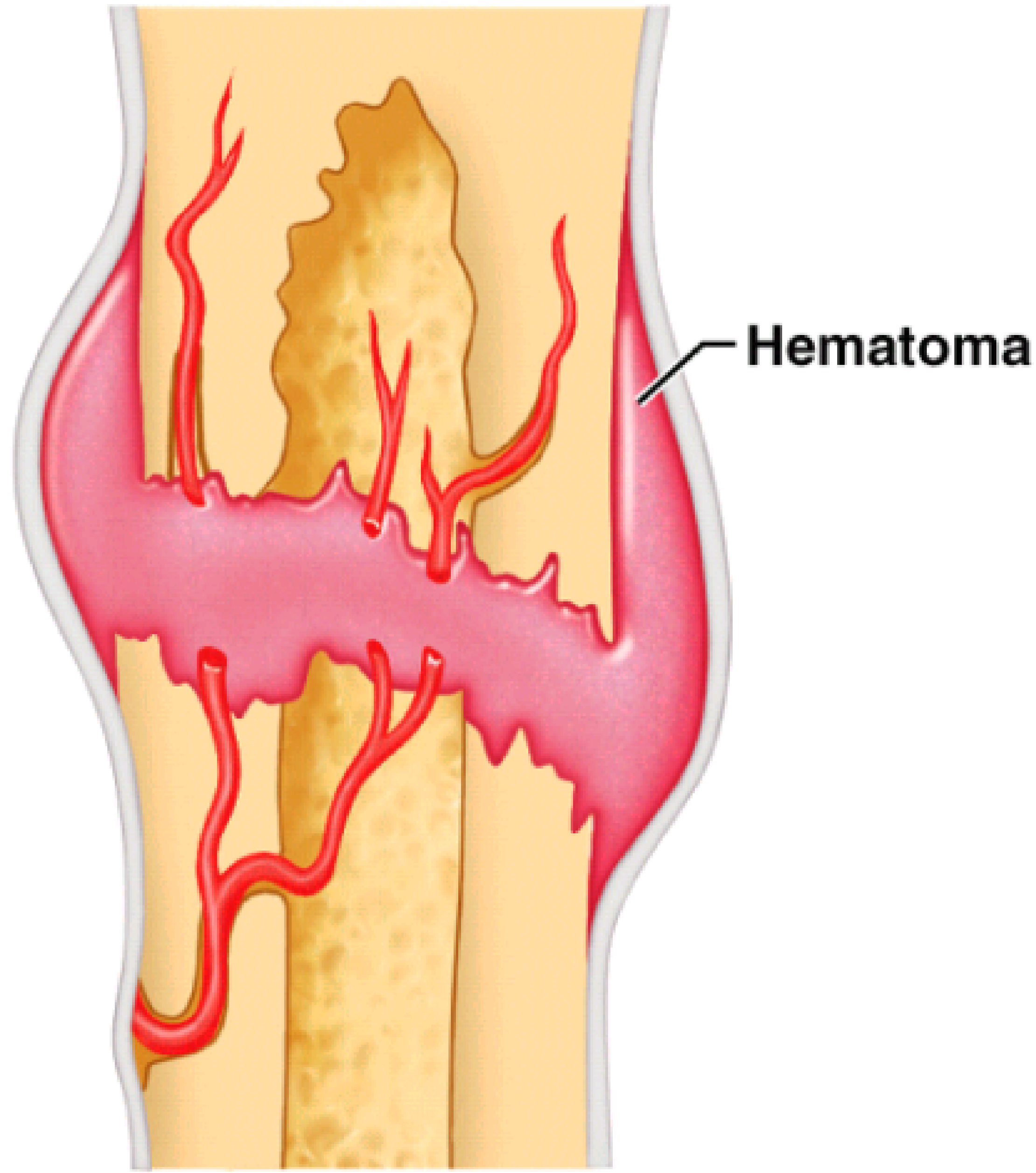
(a) Steps in appositional bone growth

Fractures

- Fractures:
  - cracks or breaks in bones
  - caused by physical stress
- Fractures are repaired in 4 steps

Fracture Repair Step 1: Hematoma

- Hematoma formation
  - Torn blood vessels hemorrhage
  - A mass of clotted blood (hematoma) forms at the fracture site
  - Site becomes swollen, painful, and inflamed
- Bone cells in the area die

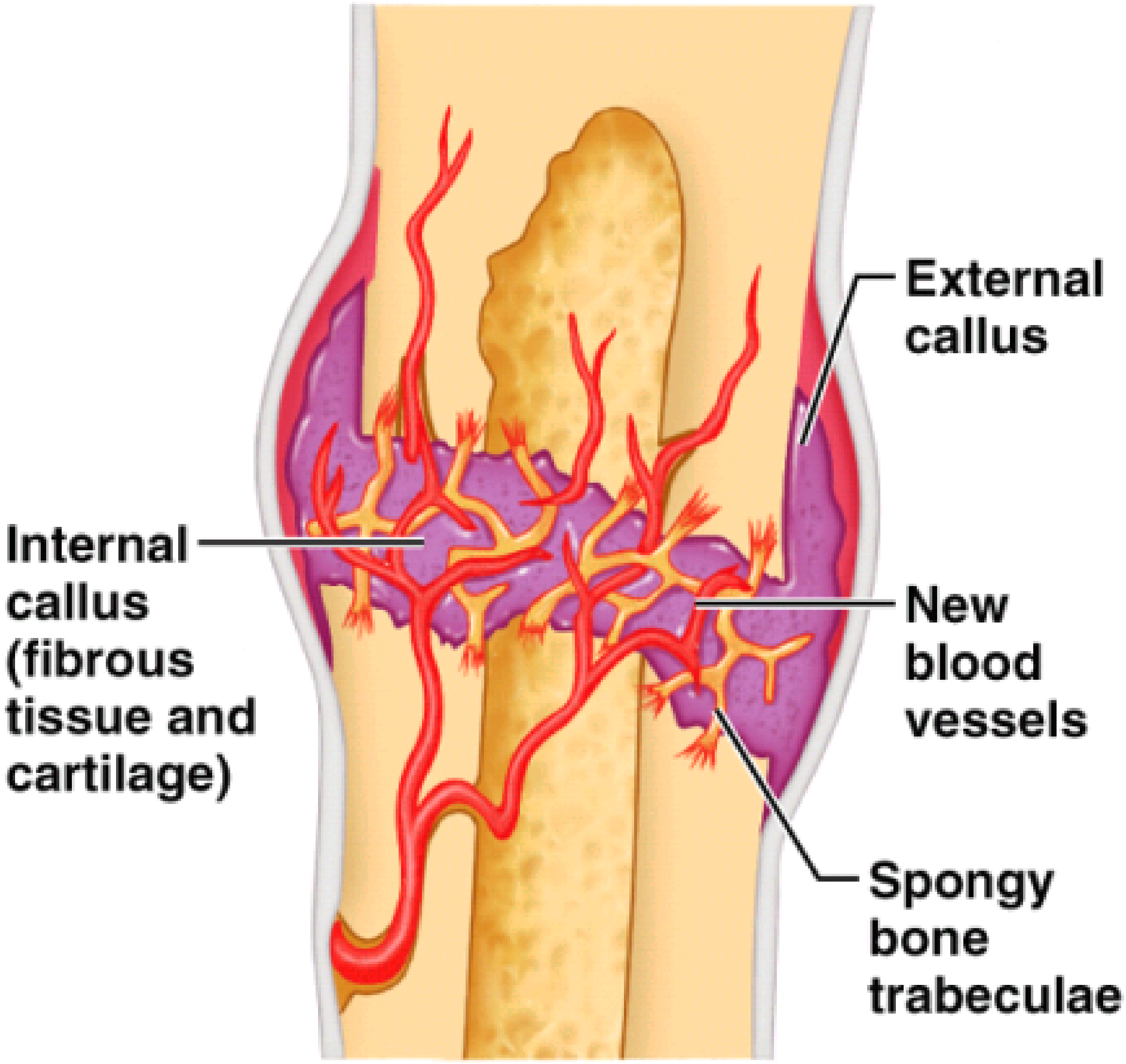


① **Hematoma formation**

Figure 6.13.1

Fracture Repair Step 2: Soft Callus

- Cells of the endosteum and periosteum divide and migrate into fracture zone
- Granulation tissue (soft callus) forms a few days after the fracture from fibroblasts and endothelium
- Fibrocartilaginous callus forms to stabilize fracture
  - external callus of hyaline cartilage surrounds break
  - internal callus of cartilage and collagen develops in marrow cavity
- Capillaries grow into the tissue and phagocytic cells begin cleaning debris



② Fibrocartilaginous callus formation

Figure 6.13.2

Fracture Repair Step 3: Bony Callus

- Bony callus formation
  - New spongy bone trabeculae appear in the fibrocartilaginous callus
  - Fibrocartilaginous callus converts into a bony (hard) callus
  - Bone callus begins 3-4 weeks after injury, and continues until firm union is formed 2-3 months later

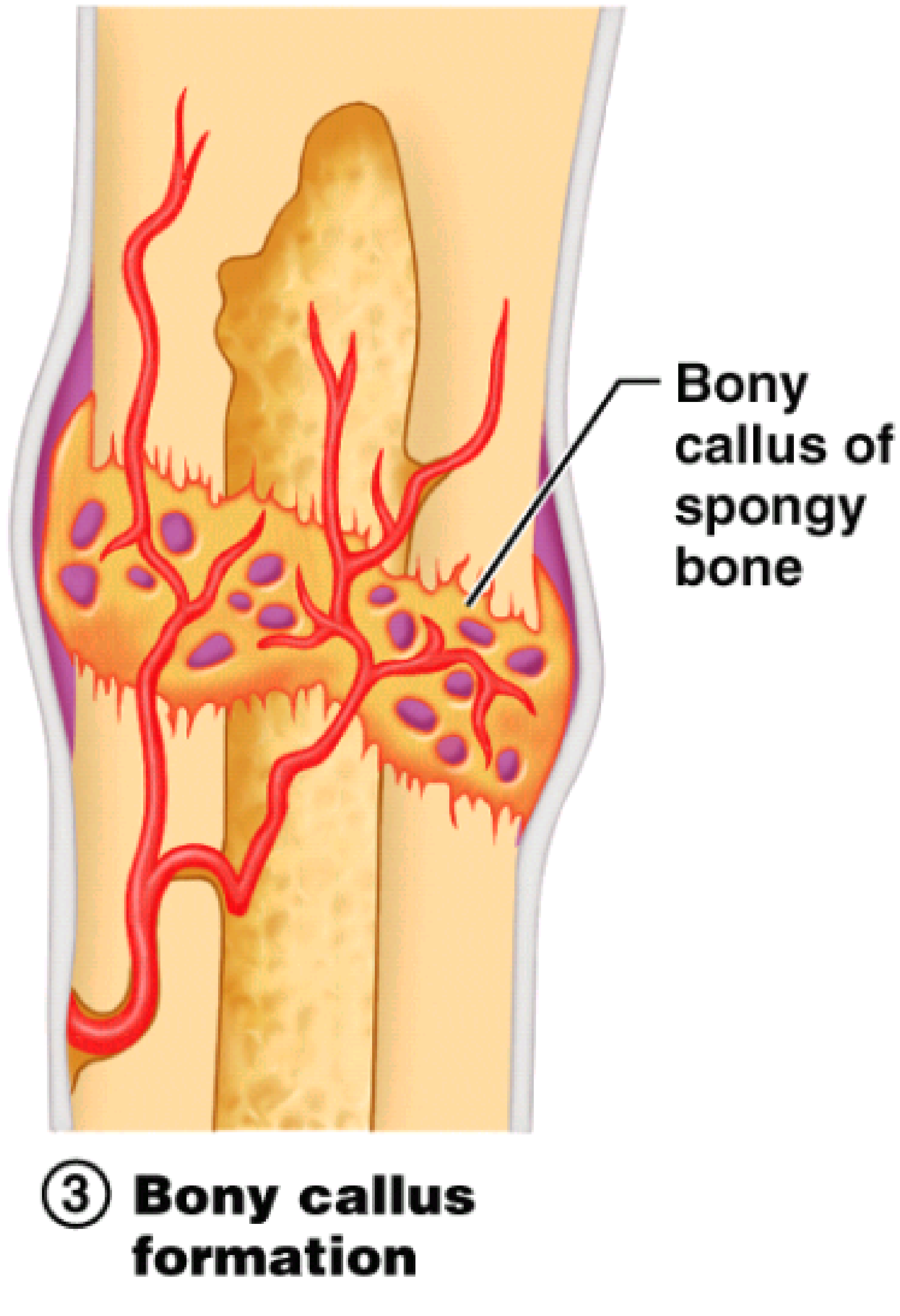


Figure 6.13.3

Fracture Repair Step 4: Remodeling

- Bone remodeling
  - Excess material on the bone shaft exterior and in the medullary canal is removed
  - Compact bone is laid down to reconstruct shaft walls
  - Remodeling for up to a year
    - reduces bone callus
    - may never go away completely
  - Usually heals stronger than surrounding bone

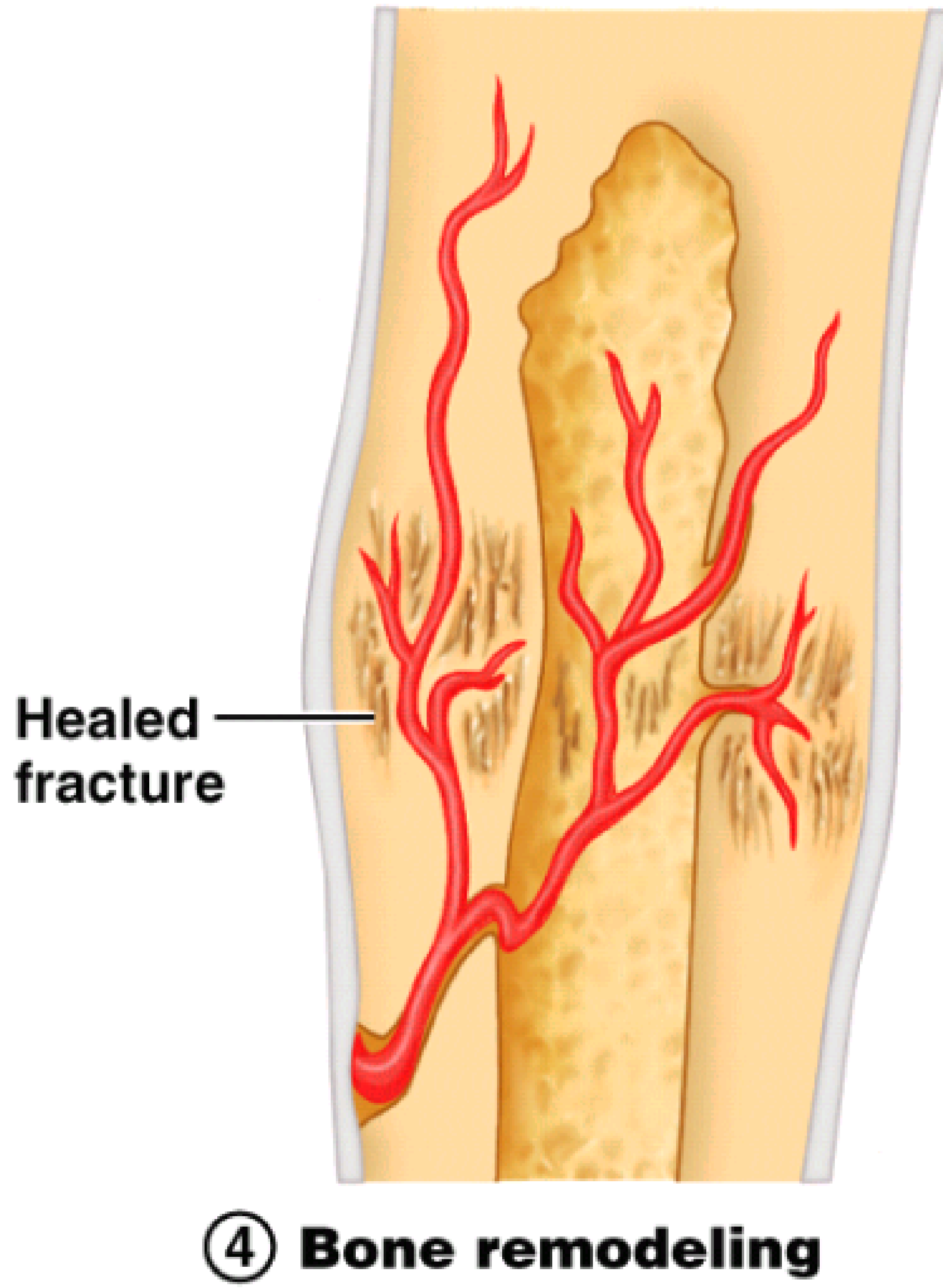


Figure 6.13.4

Aging and Bones

- Bones become thinner and weaker with age
- Osteopenia begins between ages 30 and 40
- Women lose 8% of bone mass per decade, men 3%

Hormones and Bone Loss

- Estrogens and androgens help maintain bone mass
- Bone loss in women accelerates after menopause

Cancer and Bone Loss

- Cancerous tissues release osteoclast-activating factor:
  - stimulates osteoclasts
  - produces severe osteoporosis